

EMPORIATRIC ENTERITIS: LESSONS LEARNED FROM U.S. STUDENTS IN MEXICO*,**,†

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Between 15 and 20 million persons annually cross international boundaries moving from industrialized countries to developing regions. A surprisingly large number of persons develop medical disturbances during their travels. The term "emporiatics" was created by Somerset R. Waters to encompass the various diseases of travelers. Approximately half of the persons who visit developing areas from industrialized countries experience diarrhea. The cost of diarrheal illness to the travelers is great. The cost to the countries of the developing world is even greater considering lost revenues from potential tourism and business activities which fail to develop because of the threat of developing diseases. More than 100 billion dollars are spent annually on international travel. Currently only 20% of these dollars are spent to support travel to developing areas.

The problem of travelers' diarrhea was first studied in U.S. students during short-term residence in Mexico in the 1950s and 1960s by B. H. Kean (1). Kean characterized the disease clinically, showed that conventional pathogens were not responsible and presented information that antimicrobial agents could prevent the disease. U.S. students in Mexico were largely unstudied after these investigations until 1974 when Kean joined Gorbach and provided the first clear evidence that enterotoxigenic *Escherichia coli* were responsible for a majority of the disease (2).

Based on these leads, in 1975 we began studies among U.S. students as a model for the study of travelers' diarrhea. Initially, a clinic was established at the Universidad de las Americas in rural Cholula, Puebla,

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Mexico. When a faculty strike ended school activities in 1976, we moved the research base to Guadalajara to initiate studies with the summer programs of the University of San Diego and the University of Arizona as well as the medical school, Universidad Autonoma de Guadalajara. This paper will summarize the studies which have taken place with these student populations over the last decade.

INCIDENCE OF DISEASE AND THE OCCURRENCE OF NATURAL IMMUNIZATION

Three hundred twelve asymptomatic students were enrolled in July 1975 at Universidad de las Americas (3) to determine the incidence of acute diarrhea (≥ 2 unformed stools plus one clinical indicator of enteric infection). The students were divided into three groups: newly arrived U.S. (enrolled within 24 hours of arrival in Mexico); established U.S. (present at the school one year or longer); and Latin American (from Mexico or other Latin American countries). Daily diaries were collected for one month to determine the presence of illness symptoms. When diarrhea occurred, a stool was collected for detection of enteropathogens and paired serum samples were obtained for determination of *E. coli* anti-heat-labile enterotoxin.

In Table 1, it can be seen that diarrhea occurred in a greater number of newly arrived U.S. students than the other student groups ($p < 0.005$), 40% per month compared to 20% for the established U.S. students and 12% for the students from Latin American countries. Within the Latin American students, newly arrived Venezuelan summer students did not experience diarrhea more commonly when compared to established students coming from Venezuela one year or more earlier. Recurrent episodes of diarrhea during the month were common only in the newly arrived U.S. students, occurring in 15%. Diarrhea characteristically developed in the newly arrived U.S. students during the first 16 days after

TABLE 1
*Occurrence of Diarrhea in Students at Universidad de las Americas During a One Month Study July 1975**

Student group	Number Students Studied	Number Developing Diarrhea (%)
U.S.		
Newly arrived	55	22 (40)
Established	142	28 (20)
Latin American	95	11 (12)

* Adapted from reference 3.

arrival to Mexico (mean was 10 days and median was 7 days after arrival). In this group the average number of stools passed during the first 48 hours was 13 (range 2–53) and the total consecutive days of passing unformed stools averaged 5 (range 0.5–20). The U.S. students (newly arrived and established) experienced more intense diarrhea when compared to the students from Latin America. Fifty percent of U.S. students with diarrhea passed 10 or more unformed stools in the first 48 hours of disease compared to 23% passing this number among ill Latin American students.

ETIOLOGY

Enteric pathogens were sought by accepted techniques (4, 5). In the initial study carried out at Universidad de las Americas in 1975 an enterotoxigenic *Escherichia coli* (ETEC) was isolated in diarrheal stools of 38% of the newly arrived U.S. students, in 23% for the established U.S. students and in 17% for the Latin American students (4). Using the passive immune hemolysis technique, an inverse relationship was shown between acute geometric mean *E. coli* heat labile enterotoxin (LT) antibody titer for the newly arrived U.S. students and the occurrence of diarrhea associated with LT-producing ETEC (6). Also, a significant correlation ($p < 0.005$) was found between a rise in antitoxin titer and diarrhea associated with LT-producing *E. coli*.

We have looked for ETEC and the conventional pathogens during each study during the last 10 years. Rotavirus was sought in surveys carried out during the first two years, campylobacter has been looked for during the last four years and cryptosporidium has been sought during the past three years. The relative frequency of enteropathogens found in diarrheal stools among newly arrived U.S. students with illness in these studies is shown in Table 2. Enterotoxigenic *E. coli* has been identified in approximately 40%. Shigella was found in 21%, salmonella in 7%, *Campylobacter jejuni* in 3%, *Aeromonas hydrophila* in 1%, and *Giardia* in

TABLE 2
Identification Rates of Enteric Pathogens in U.S. Students Newly Arrived in Mexico

Enteropathogen	Percent Found
ETEC	43%
Shigella	21%
Salmonella	7%
Campylobacter	3%
Aeromonas	1%
Giardia	2%
Unknown	23%

2%. In approximately 23% an established etiologic agent has not been found. During the past two years, we have provided evidence that in up to 15% of newly arrived U.S. students with illness or approximately 30% of the unknown cases, a nonenterotoxigenic HEp 2 cell-adherent *E. coli* strain could be recovered (7). Further evidence of the pathogenicity of this new type of organism, resembling classical enteropathogenic *E. coli* in its enteroadherence properties, was provided by showing disease-producing capability when a strain was fed to adult volunteers (8). In two years of study we have found cryptosporidium only rarely in this population. Rotavirus, while commonly found in diarrheal stools, was also detected frequently in students without symptoms (9, 10). Also, bacterial agents were commonly found in rotavirus-positive diarrheal stools.

FOOD AND WATER

Forty-nine of the 55 newly arrived U.S. students in the initial cohort study looking at incidence of illness completed a daily questionnaire detailing place of food consumption and specific food items consumed for a one month period of time (11). Among the newly arrived U.S. students who ate half or more of their meals in public restaurants and the school cafeteria, there were significant increases in diarrhea ($p < 0.005$), shigellosis ($p < 0.05$) and ETEC diarrhea ($p < 0.025$) compared to the students who self-prepared half or more of their meals in their own apartments (Table 3). Although not shown in Table 3, there was also a statistical association with food consumption and diarrhea for the subgroup eating more than the average from street vendors. In this earlier study foods were sampled and cultured qualitatively for enteric pathogens. High numbers of enteric bacteria were recovered from food obtained from the school cafeteria, public restaurants, street vendors and grocery stores. *Shigella* was isolated from cooked and uncooked ham-

TABLE 3
*Occurrence of Diarrheal Illness, Shigellosis and Enterotoxigenic E. coli Diarrhea Among 49 Newly Arrived U.S. Students (Universidad de las Americas) July 1985 According to Location by Food Consumption**

Location ≥50% of Meals	Number Students	Diarrhea	Shigellosis	ETEC Diarrhea
Private				
Apartments	20	3 (15%)	1 (5%)	0
Public				
Restaurants and/or school cafeteria	29	13 (45%)	9 (31%)	10 (34%)

* Adapted from reference 11.

burger patties from the school cafeteria. Four shigella carriers were identified from among the food handling kitchen personnel at the school.

Soon after moving our research unit to Guadalajara we continued the study of food quality (12). In this urban area, we also had the opportunity to study the effect of eating food in the homes of Mexican families participating in the student housing programs. These represented middle class homes with refrigerators, telephones, televisions and live-in household help. In Table 4 the frequency of consumption of foods prepared in Mexican homes can be seen to be highly associated with the greater risk of acquisition of diarrhea. Foods from commercial sources and private Mexican homes in Guadalajara were subsequently examined for contamination with coliforms, fecal coliforms and bacterial enteropathogens (12). For comparison, selected restaurant foods were obtained in Houston (from Mexican restaurants). The data obtained are presented in Tables 5 and 6. The foods from Mexican sources contained a mean of between 800 and 3,000 coliforms/g while these foods yielded between 200 and 900 mean fecal coliforms/g. Of interest, the 12 Houston foods contained impressive numbers of coliforms and fecal coliforms although somewhat

TABLE 4
*Association of Frequency of Consuming Foods in Mexican Homes and Development of Diarrhea, U.S. Students in Guadalajara, Mexico During the Summer of 1980**

	Percent of Food Consumed in Mexican Homes		
	0-39	40-79	≥80
Number of Students	22	84	31
Number Developing Diarrhea	1	32	16
Percent Ill	(5%)	(38%)	(52%)

* Adapted from reference 12.

TABLE 5
*Isolation of Coliforms and Fecal Coliforms From Selected Supermarkets and Public Eating Establishments in Guadalajara and Houston**

Source	Number of Foods	Coliforms (Mean)	Fecal Coliforms (Mean)
Mexico			
Supermarket	5	3,349	529
Cafeteria	5	1,108	940
Street Vendors	5	844	232
Restaurants	11	828	487
Houston	12	561	189

* Adapted from reference 12.

TABLE 6
*Characterization of Fecal Coliforms Obtained from Foods in Mexico and Houston—Occurrence of E. coli, Enterotoxigenic E. coli and Other Enteric Pathogens**

Source	Number of Isolates	Number of <i>E. coli</i>	Number of ETEC	Number Other Pathogens
Mexico				
Restaurants	162	40	4	0
Homes	353	72	6	22
Houston				
Restaurants	30	0	0	0

* Adapted from reference 12.

less in number. When the fecal coliforms were further studied and enteric pathogens sought more revealing information was developed (Table 6). The foods from Guadalajara, both from restaurants and Mexican homes commonly contained *E. coli* and less commonly enterotoxigenic *E. coli*. Neither was found in the foods studied in Houston. The 22 other pathogens isolated from foods obtained in Mexican homes consisted of 17 salmonella, 4 shigella and 1 *Aeromonas hydrophila*.

In two surveys rotavirus infection was sought by direct visualization of the agent in stool using the electron microscope. Rotavirus infection rates were not found to correlate with food consumption patterns suggesting that a non-food source might be responsible for this enteric infection (10). In 1978 (13) and in 1982 (14) water samples were collected for study from various sources in Guadalajara, the Lake Chapala area and Puerto Vallarta. Twenty liter samples were subjected to filter and adsorption-elution techniques for detection of enteric viruses. Rotavirus was sought by plating the filtered material on fetal rhesus monkey kidney cells (MA 104 cells) and examined for infectious fluorescent foci by indirect immunofluorescence. Enteroviruses were detected by standard techniques.

In the initial survey, carried out during the rainy season of August 1978, 10 of 10 drinking water samples contained viable rotavirus and Coxsackie B4, B5 or B6 were found in 5 of 10 (Table 7). When the studies were repeated during the dry season of December 1979, rotavirus was recovered from 3 and enteroviruses from 8 of 21 drinking water samples. Water quality data indicated that while many tapwater samples did not meet U.S. coliform standards, some samples containing infectious viruses did. In the second study, carried out in the two different seasons in 1982, similar results were obtained (14). This second study showed the failure of water treatment facilities in Guadalajara to remove enteric viruses from treated water. Again, water contamination by coliforms and enteric viruses was most pronounced during the rainy season.

TABLE 7
*Identification of Viable Rotaviruses and Enteroviruses in Public Drinking Water in Mexico 1978-1979**

Season	Number Specimens Examined	Rotavirus Foci/20 l water		Enteroviruses/ 20 l water
		Number Positive (%)	Range (Mean)	Number Positive (%)
Rainy August, 1978	10	10 (100)	16-210 (103)	5** (50)
Dry December, 1979	21	3 (14)	37-2500 (883)	8† (38)

* Adapted from reference 13.

** Coxsackie B4, B5 and B6

† Coxsackie B3

CHEMOTHERAPY AND CHEMOPROPHYLAXIS

We have carried out a number of studies of the treatment and prevention of travelers' diarrhea in the U.S. student population. These studies have largely established the current recommendations for therapy of acute travelers' diarrhea (15). Bismuth subsalicylate (16) and loperamide (17) have been shown to effectively reduce the symptoms associated with the illness. Trimethoprim/sulfamethoxazole (TMP/SMX) and trimethoprim alone have been used as both prevention and therapy of travelers' diarrhea as seen in our students (18, 19). These studies will be briefly summarized.

Within 48 hours of arrival in Guadalajara during the summer of 1980, 57 students were given TMP/SMX (160 mg TMP/800 mg SMX), 58 were given TMP (200 mg) and 30 were given a placebo once daily for 14 days to determine their protective effects (18). Diarrhea, defined as ≥ 4 unformed stools/24 hours, or 3 in 8 hours plus one or more clinical indicators of enteric infection (cramps, pain, fever, nausea, vomiting) occurred in 1 of 57 of those taking TMP/SMX, 8 of 58 taking TMP alone and 10 of 30 taking a placebo. This gave an overall protection rate of TMP/SMX of 95%; for TMP it was 52%. One of those receiving TMP/SMX (2%) and two of those taking TMP (3%) experienced a self-limiting skin eruption. All receiving active drug transiently developed a totally antimicrobial resistant aerobic gut flora during prophylaxis (20). The common finding of TMP/SMX resistant coliforms in foods obtained in Mexico (21) probably explains this finding. Although a recent Consensus Conference panel did not recommend the use of prophylactic antibiotics in controlling travelers' diarrhea (15), we feel that with further study it may be possible to identify host and destination factors which

could be used in selecting a subset of persons who might be advised to use this approach.

In a separate therapeutic trial carried out during the summer of 1981, the same two drugs were used in double the daily dose, or 160 mg TMP/800 mg SMX or TMP 200 mg each taken twice daily for 5 days (19). Thirty-seven students with illness (defined as in the prophylaxis trial) received TMP/SMX, 39 TMP alone and 35 a placebo. Mean duration of diarrhea, as defined as the time from administration of medication until the last unformed stool was passed, was 29 hours for TMP/SMX, 31 hours for TMP and 93 hours for placebo. Statistical reductions in diarrhea were seen for illness due to ETEC and shigella strains as well as for illness unassociated with a detectable agent. The drugs were well tolerated. One student receiving TMP alone (3%) developed a self-limiting skin rash.

SUMMARY

In the studies reported, evidence has been presented that U.S. students traveling to Mexico represent a model for the study of travelers' diarrhea. The incidence of illness acquisition approximates that published in other studies of travelers (22). Natural immunity was shown to develop as students remained in Mexico presumably through repeated exposure to prevalent agents, particularly ETEC. ETEC, shigella strains and no detectable agent represented the largest groups when etiologic assessment was made. Food probably served as the important source of diarrhea particularly that due to ETEC and shigella strains. The level of bacteria isolated from food suggested that organism replication occurred due to improper temperature storage rather than to heavy initial contamination. The location of food consumption was related to degree of risk: self preparation was the safest, eating in Mexican homes the least safe and consumption of food in public restaurants was intermediate in risk. Water probably played a role in the transmission of viral infection. The risk of water contamination appeared to be highest during the rainy seasons. Finally, the antimicrobial agents TMP/SMX and TMP alone were shown to effectively prevent and treat this form of travelers' diarrhea.

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DISCUSSION

Woodward (Baltimore): Dr. DuPont, that was up to your usual high standard of presentation. Thank you. Would you say a word about the virtues, or lack of them, regarding the taking of antiperistaltics?

DuPont: Well, Dr. Woodward is asking an important question. In 1973 Dick Hornick and I published a paper in the *Journal of the American Medical Association* showing that occasionally febrile dysenteric shigellosis can be made worse by taking diphenoxylate HCl (Lomotil). Robert Steffen additionally has performed a study in Swiss and German travelers looking at the prophylactic potential of the active metabolite of diphenoxylate in the prevention of travelers' diarrhea. Interestingly, he demonstrated that a significantly greater number of tourists who took the active drug (compared to placebo-treated individuals) acquired diarrhea. He had created the experimental animal model of enteric infection. A laboratory animal must be pre-treated with opiate-like drugs to paralyze the intestine before infection by an enteric organism can result. We have continued these studies and feel that antimotility drugs can make certain forms of gastroenteritis worse, particularly febrile dysenteric diseases. But, on the other hand, for non-febrile cases of diarrhea where there is no blood being passed (absence of dysentery), these drugs do not appear to make the disease worse. At least we have not demonstrated a potentiation of the illness. We currently believe these drugs can be used in non-febrile, non-dysentery cases of disease but they should not be continued longer than 36 hours and should be discontinued in all cases not improved by the therapy.

Bransome (Augusta): There's an apparent association, occasionally anyway, of sero-negative polyarthritis following some of these infections. Would you comment on that?

DuPont: Yes. Reiter's syndrome type of disease has been associated with *Shigella flexneri* infection in association with the histocompatibility antigen HL-A B27. Thus the genetics of the individuals and the specific strains of the bacteria both appear to be involved in development of the syndrome.

Thorup (Charlottesville): A number of years ago, while living in Tucson, we were struck with the number of Mexican visitors who came to Tucson and had the same kinds of problems we had when we went to Mexico. I wonder if you've repeated these studies with Mexican students who come up here and live in various parts of this country for periods of time.

DuPont: This is a statement or concept that I have heard frequently. There are two studies that have been carried out to look at the occurrence of diarrhea in Mexican students coming to the United States; one done in Miami, another in Southern California. Both of these studies have indicated that diarrhea does occur when Mexican students come to the United States but it is unusual and clinically very mild. I will say, when we looked at our students with diarrhea and divided them into two groups, very mild diarrhea and severe diarrhea, we found that the mild diarrhea was generally not associated with a pathogen and the severe diarrhea was associated with a pathogen in over 90% of the cases. So we believe that mild diarrhea of travelers—which can explain the illness of your Mexican students coming to Tucson—probably is not infectious in origin. This mild symptomatology may relate to dietary differences, alcohol consumption patterns or to psychic stresses. The severe disease that leads to bed confinement is not the kind of thing we normally see in persons traveling from high risk to low risk areas. This striking illness is characteristic among people traveling from low risk areas (U.S., Canada, Western Europe) to high risk areas (Latin America, Asia, Africa) and is due to an infectious microorganism.

Schreiner (Washington): I was fascinated by your numbers on food preparation at home versus restaurants. I wonder if you would hazard a speculation. Do you think this contamination actually occurs in the home or is it a difference in the source of the food? I would presume that a restaurant, for example, certainly in the northern part of the U.S., might purchase its raw materials from a totally different source, than a Mexican family which might be buying in a market that is subject to animal contamination and so forth. Do you think this increased risk of contaminants is something that occurs in the home because it's put into the food in the home, or do you think it's a difference in the source of foods coming into the home?

DuPont: That is a good question. I think the answer is: "all of the above". The food is contaminated at all levels; almost every error in food hygiene is made in these parts of the world. While food in the super-market in Mexico is contaminated to begin with, I think the most important single contributor is inadequate preservation of food at low temperatures in the homes after preparation. The levels of bacterial counts that we found must have occurred through active bacterial replication in the food. It probably is not initial contamination alone.

Warren (Columbus): I was going to ask the same question—why the home was so bad and you've answered that. But one facet of it I'd still like to ask. What about the role of alcohol. There is an awful lot of beer drunk in Mexico and does that play a role in this difference that we see.

DuPont: Since I had 12 minutes to present ten years of data, I did not talk about the specific food items that we have cultured or studied in other ways. Evidence would suggest that carbonated soft drinks including beer are safe. We published a paper last May in the *Journal of the American Medical Association* looking at the influence of various popular drinks on bacterially contaminated ice. We were interested in whether booze might decontaminate ice. Many of us are careful not to drink water in places where we travel, but when we have a drink in the evening we tend to put an ice cube or two in the drink hoping that the alcohol or carbonated soft drink will decontaminate the ice. I have got mixed news for you. If you take a mixed drink like scotch and soda, there is not enough alcohol in that to do much good if you have contaminated ice; if you use straight 86 proof tequila, you can cut the organisms down about 90%, which might be clinically important for preventing infection by non-shigella enteric organisms. Soft drinks (club soda and colas) had minimal effect on the survival of enteropathogens in ice. These studies would suggest that it is probably wise to resist the temptation to use ice in most tropical areas of the developing world.